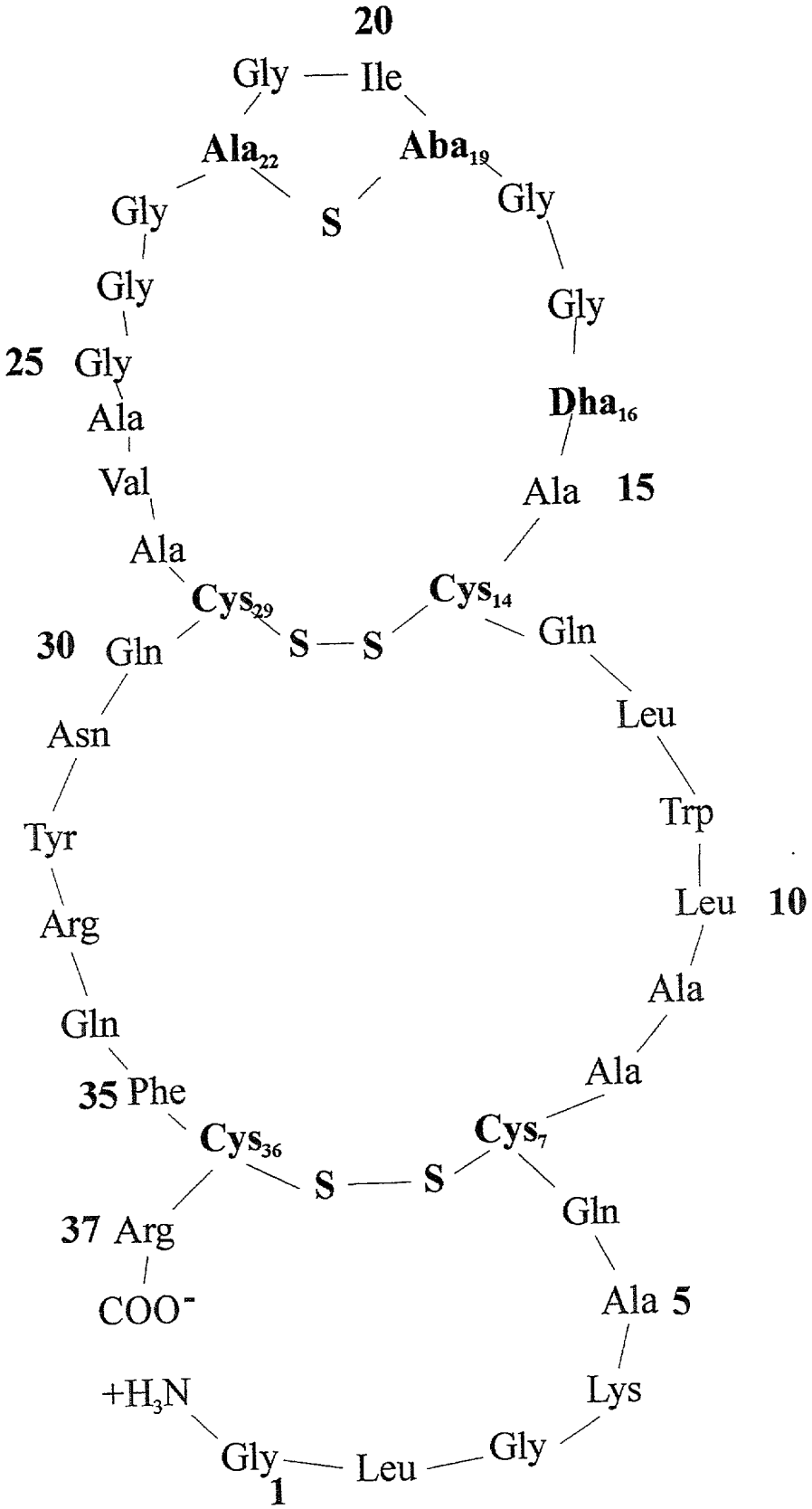


Sublancin 168 **Figure 1**

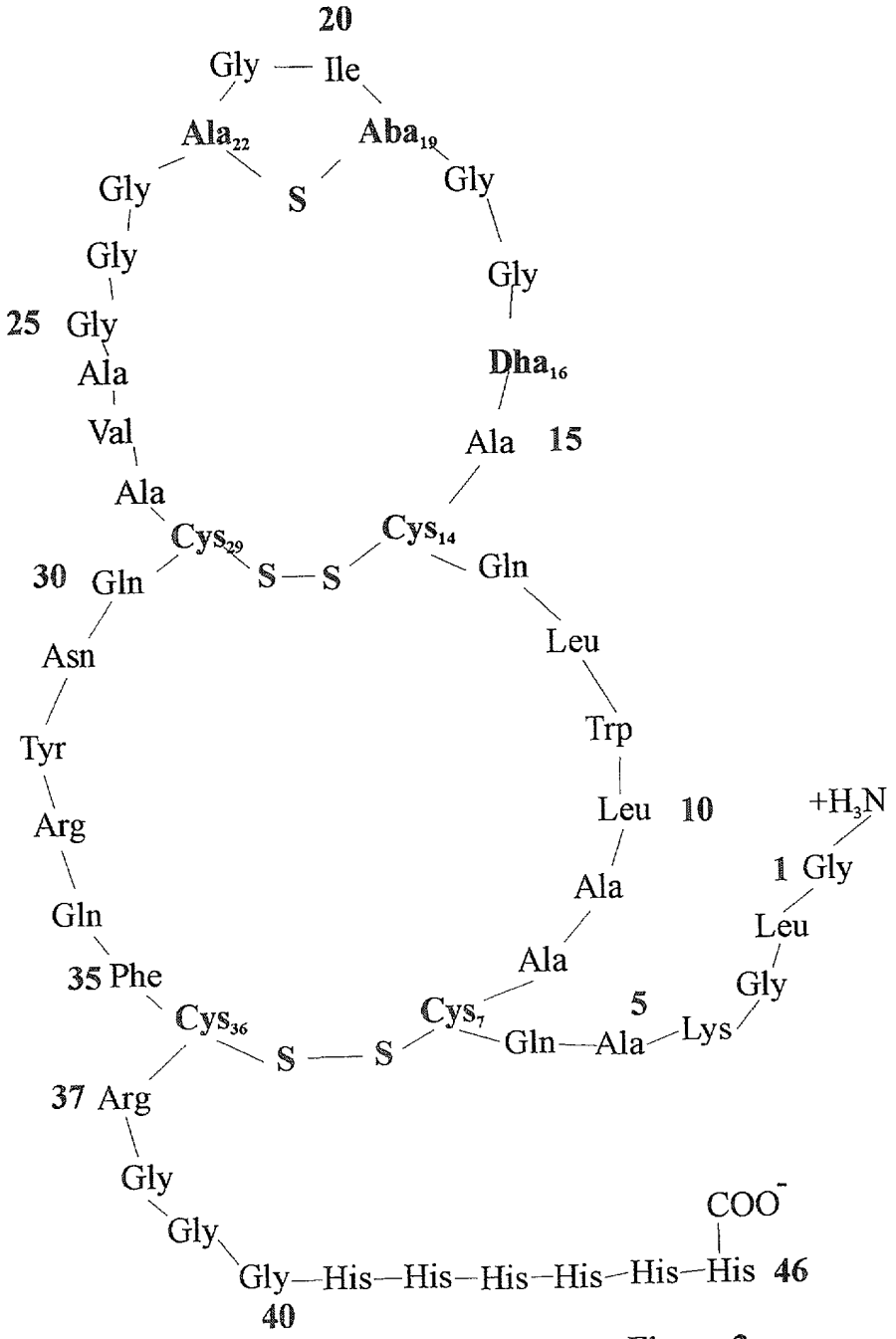
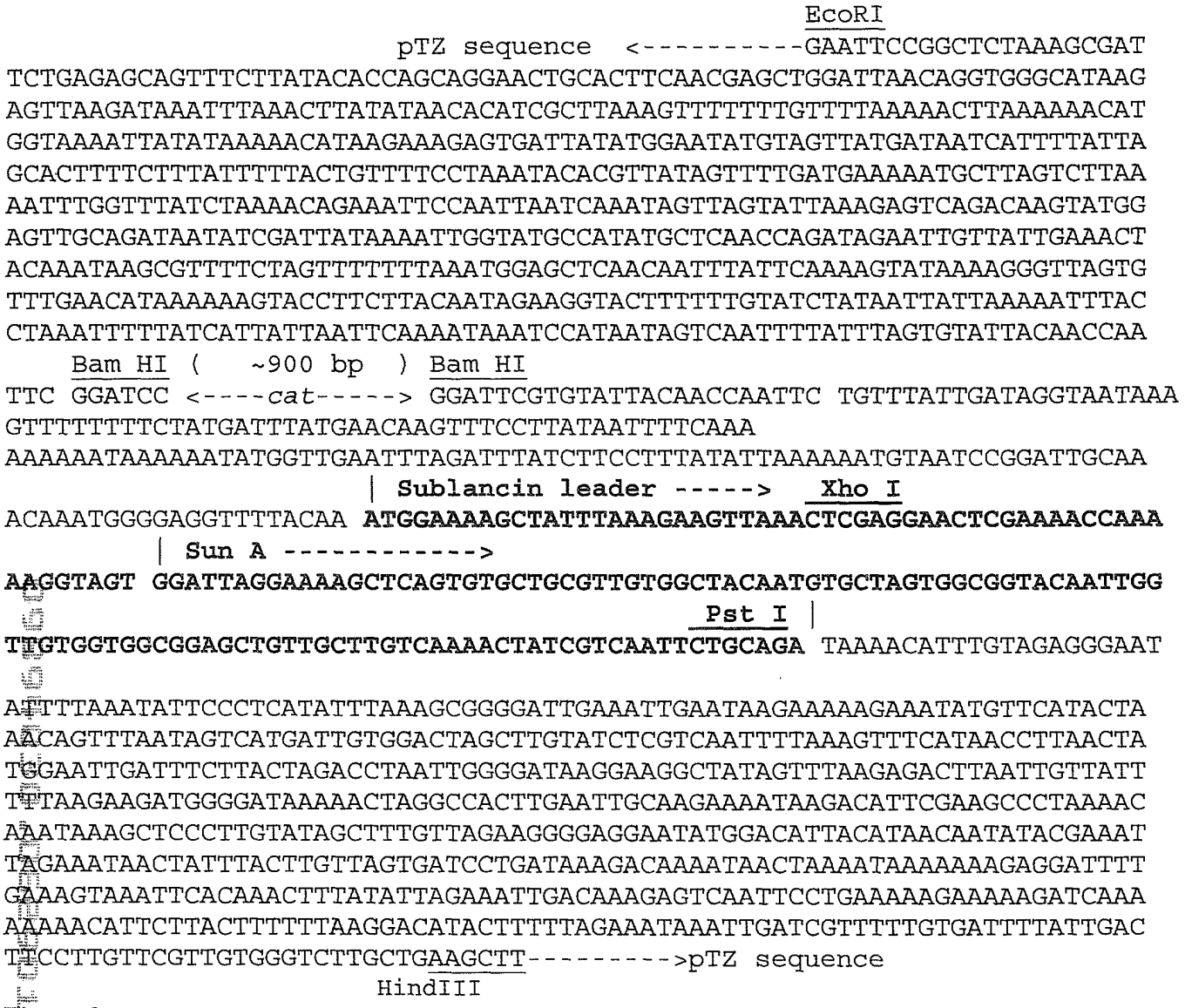


Figure 2

Sublancin-His tag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GGTAAAATTATATAAAAACATAAGAAAGAGTGATTATATGGAATATGTAGTTATGATAATCATTTTATTA
GCACTTTTCTTTATTTTTACTGTTTTCCTAAATACACGTTATAGTTTTGATGAAAAATGCTTAGTCTTAA
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AGTTGCAGATAATATCGATTATAAAATTGGTATGCCATATGCTCAACCAGATAGAATTGTTATTGAAACT
ACAAATAAGCGTTTTCTAGTTTTTTTAAATGGAGCTCAACAATTTATTCAAAAGTATAAAAGGGTTAGTG
TTTGAACATAAAAAAGTACCTTCTTACAATAGAAGGTACTTTTTTGTATCTATAATTATTAAAAATTTAC
CTAAATTTTTATCATTATTAATTCAAAATAAATCCATAATAGTCAATTTTATTTAGTGTATTACAACCAA
Bam HI (~900 bp) Bam HI
TTC GGATCC <----cat-----> GGATTCGTGTATTACAACCAATTC TGTTTATTGATAGGTAATAAA
GTTTTTTTTCTATGATTTATGAACAAGTTTCCTTATAATTTTCAAA
AAAAAATAAAAAATATGGTTGAATTTAGATTTATCTTCCTTTATATTAAAAAATGTAATCCGGATTGCAA
| Sublancin leader -----> Xho I
ACAAATGGGGAGGTTTTACAA ATGGAAAAGCTATTTAAAGAAGTTAAACTCGAGGAACTCGAAAACCAAA
| Sun A ------------>
AAGGTAGT GGATTAGGAAAAGCTCAGTGTGCTGCGTTGTGGCTACAATGTGCTAGTGGCGGTACAATTGG
Pst I |
TTGTGGTGGCGGAGCTGTTGCTTGTCAAAACTATCGTCAATTCTGCAGA TAAAACATTTGTAGAGGGAAT
ATTTTAAATATTCCCTCATATTTAAAGCGGGGATTGAAATTGAATAAGAAAAAGAAATATGTTCATACTA
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GAAAGTAAATTCACAAACTTTATATTAGAAATTGACAAAGAGTCAATTCCTGAAAAAGAAAAAGATCAAA
AAAAACATTCTTACTTTTTTAAGGACATACTTTTTAGAAATAAATTGATCGTTTTTGTGATTTTATTGAC
TTCCTTGTTCGTTGTGGGTCTTGCTGAAGCTT---------->pTZ sequence
HindIII

Figure 3

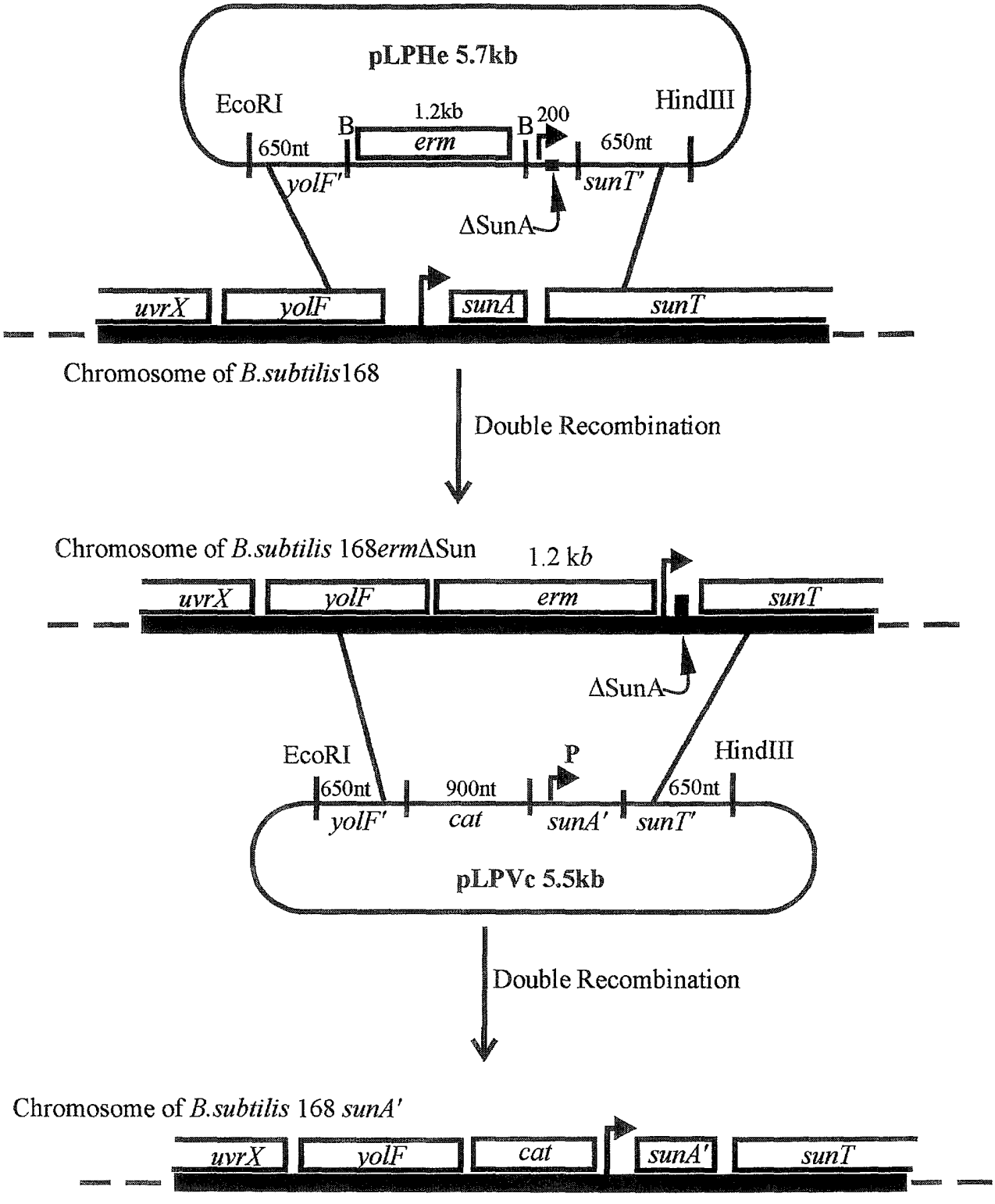

pLPHe 5.7kb
EcoRI
1.2kb
B
B
200
HindIII
650nt
erm
650nt
yolF'
sunT'
ΔSunA
uvrX
yolF
sunA
sunT
Chromosome of B.subtilis168
Double Recombination
Chromosome of B.subtilis 168ermΔSun
1.2 kb
uvrX
yolF
erm
sunT
ΔSunA
EcoRI
P
HindIII
650nt
900nt
650nt
yolF'
cat
sunA'
sunT'
pLPVc 5.5kb
Double Recombination
Chromosome of B.subtilis 168 sunA'
uvrX
yolF
cat
sunA'
sunT

Figure 4

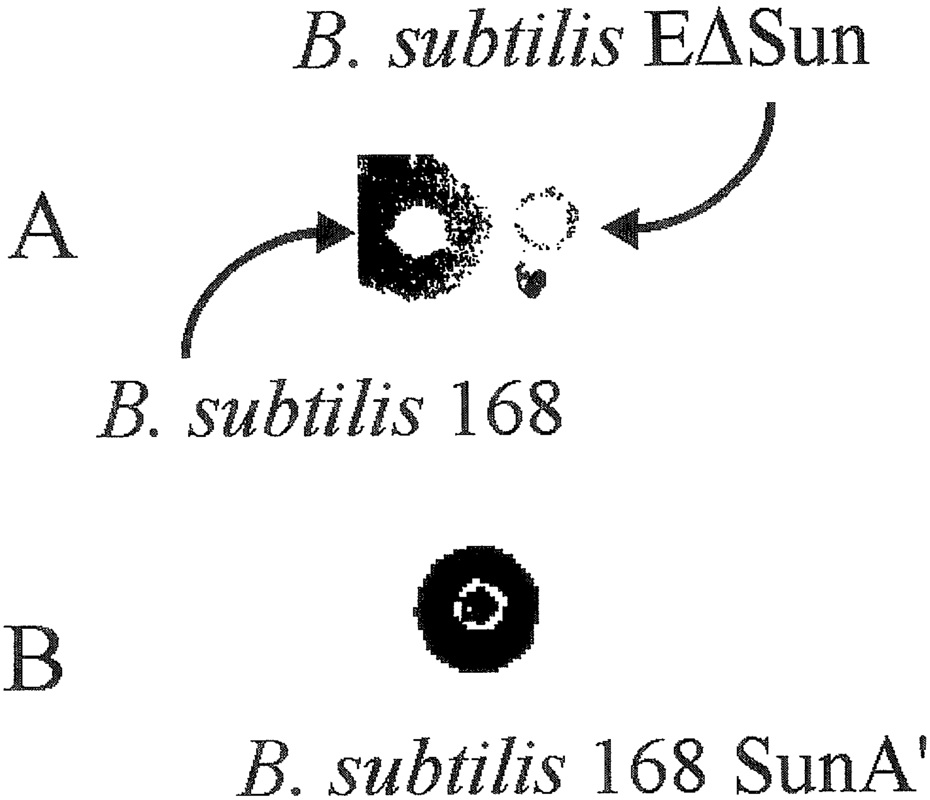
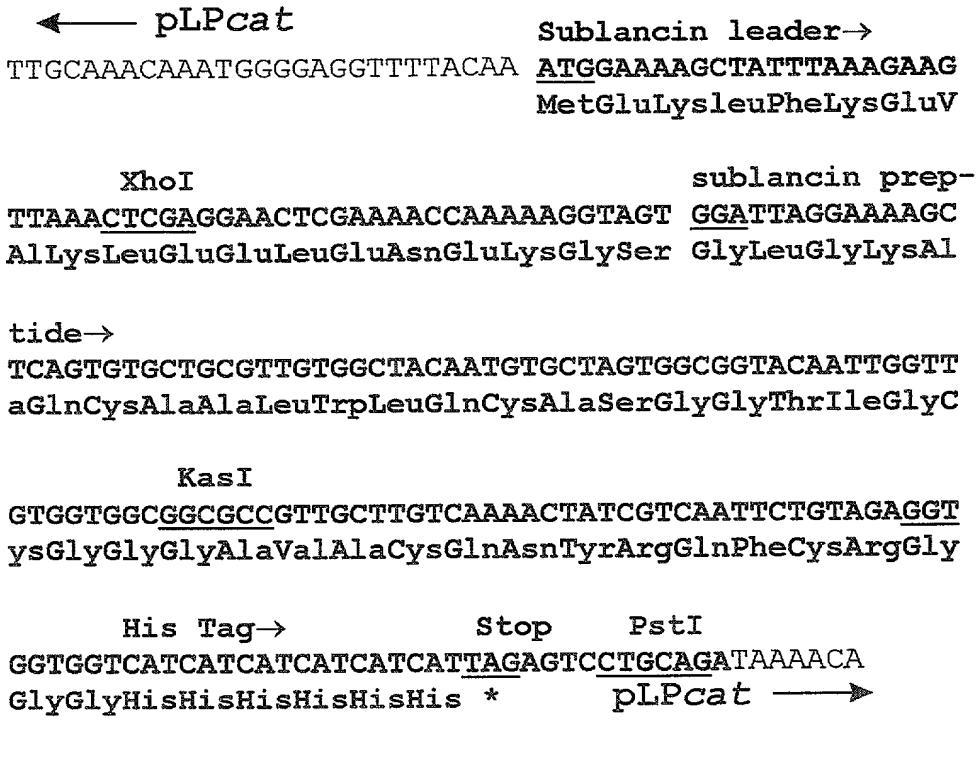


Figure 5


← pLPcat
Sublancin leader→
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MetGluLysleuPheLysGluV
XhoI
sublancin prep-
TTAAACTCGAGGAACTCGAAAACCAAAAAGGTAGT GGATTAGGAAAAGC
AlLysLeuGluGluLeuGluAsnGluLysGlySer GlyLeuGlyLysAl
tide→
TCAGTGTGCTGCGTTGTGGCTACAATGTGCTAGTGGCGGTACAATTGGTT
aGlnCysAlaAlaLeuTrpLeuGlnCysAlaSerGlyGlyThrIleGlyC
KasI
GTGGTGGCGGCGCCGTTGCTTGTCAAAACTATCGTCAATTCTGTAGAGGT
ysGlyGlyGlyAlaValAlaCysGlnAsnTyrArgGlnPheCysArgGly
His Tag→
Stop
PstI
GGTGGTCATCATCATCATCATCATTAGAGTCCTGCAGATAAAACA
GlyGlyHisHisHisHisHisHis *
pLPcat →

Figure 6